International Journal of General Medicine

Dovepress
open access to scientific and medical research

Open Access Full Text Article

SHORT REPORT

A Comparison of the Accuracy of Handheld Hemoglobinometer and Hematocrit Measurements for Detecting Plasma Leakage in Dengue Hemorrhagic Fever

Silvita Fitri Riswari[1,2], Muhammad Fitriandi Budiman[2,3], Dera Darmayanti[2], Ernawati[2], Susantina Prodjosoewojo[2,3], Evan Susandi[2,3], Amaylia Oehadian[3], Bachti Alisjahbana[2,3]

[1]Department of Biomedical Sciences, Faculty of Medicine, Universitas Padjadjaran, Bandung, Indonesia; [2]Research Center for Care and Control of Infectious Diseases, Universitas Padjadjaran, Bandung, Indonesia; [3]Department of Internal Medicine, Hasan Sadikin General Hospital, Bandung, Indonesia

Correspondence: Bachti Alisjahbana, Tel +62 823 9841 5612, Fax +62 22 204 4128, Email b.alisjahbana@gmail.com

Purpose: Practical methods for detecting plasma leakage should be readily available in all areas where dengue is endemic. We compared the accuracy of measurements obtained with a handheld HemoCue® Hb 201 instrument used for hemoglobin point-of-care testing (Hb-POCT) with that of measurements of hematocrit (Ht) levels for detecting plasma leakage in dengue patients.

Patients and Methods: We performed both measurements using the HemoCue® Hb201 system and microhematocrit method on EDTA blood taken from dengue patients at three time points during their hospitalization. Ascites, pleural effusion, or gallbladder thickening determined through ultrasound examinations were considered the gold standard for determining dengue hemorrhagic fever (DHF) versus dengue fever (DF).

Results: Close agreement between Hb-POCT and Ht measurements was indicated by an r square value of 0.845 in a linear regression. The sensitivity results for distinguishing between DHF and DF at admission were similar for Hb-POCT (63.6%) and Ht (66.7%) (Kappa = 0.75) using the optimal cutoff point determined via ROC analysis. Delta differences (in percentage) for Hb-POCT and Ht between the highest and lowest values showed lower sensitivity (45.5% and 48.5%, respectively; Kappa 0.60) when the optimal cutoff point was applied. Recommended cutoffs of ≥20% to confirm plasma leakage provided a slightly higher sensitivity using Hb-POCT (18.2%) compared with the sensitivity obtained using Ht (15.2%) with Kappa value of 97.9%.

Conclusion: Our results showed that the accuracy of Hb POCT measurements was similar and not inferior to Ht measurements for detecting plasma leakage in patients with DHF. We recommend that further evaluations are conducted to determine the optimal cutoff point given the low sensitivity associated with using ≥20% Hb-POCT or Ht increases to determine hemoconcentration.

Keywords: DHF, HemoCue®, hemoglobin, sensitivity, hemoconcentration

Introduction

Although dengue is one of the most important arboviral diseases in the world,[1] its pathophysiology is not well understood.[2] Clinical manifestations vary from mild common cold-like illness to severe dengue hemorrhagic fever (DHF) and life-threatening dengue shock syndrome caused by plasma leakage. Temporary dysfunction of endothelial cells in dengue infection increases vascular permeability, releasing plasma into extracellular spaces. This phenomenon is evidenced by increased concentrations of blood cellular components measured by hematocrit (Ht) levels. In severe cases, plasma volume decreases by more than 20% and pleural effusion and hypoproteinemia occur.[3,4]

The primary management strategy of dengue fever (DF) entails early detection of plasma leakage and fluid replacement for patients in hypovolemic states. It is well established that increased Ht levels indicate plasma leakage, prompting patient referrals for fluid replacement therapy.[5] Given the importance of Ht measurements in decisions on the

Received: 23 October 2021
Accepted: 13 January 2022
Published: 5 March 2022

use of this therapy, this test should be readily available in all primary health-care clinics that are accessible to communities in countries where dengue is endemic. However, Ht tests require a well-running centrifuge or a sophisticated hematology analyzer and are not always available in primary health clinics.

Hemoglobin (Hb) measurements usually reveal the presence of anemia. Logically, like Ht, they also reflect red blood cell concentrations. Hb and Ht are reported to be significantly correlated.[6] Moreover, many more affordable mobile options for measuring Hb are available, such as the HemoCue® Portable Photometer. HemoCue® is a Hb point-of-care testing (Hb-POCT) instrument that is available in many remote health centers and can be used in a patient's home. It is one of the most widely used instruments for measuring Hb in resource-limited settings, notably in mother-child health programs and blood donation centers.[7,8] Studies have reported satisfactory results that are consonant with those obtained using automated hematology analyzers as well as adequate inter- and intra-instrument reliability.[9,10] Nevertheless, Hemocue® and the usefulness of Hb as an indicator are not mentioned in the WHO guidelines for treating dengue.[5] Therefore, we aimed to determine whether Hb testing using HemoCue® demonstrated a good correlation and was not inferior to Ht testing for detecting hemoconcentration or plasma leakage in dengue patients.

Materials and Methods

We conducted a prospective diagnostic assessment from January to June 2015 in three hospitals in Bandung, Indonesia. Eligible subjects were hospitalized patients aged 14 years or above with a dengue diagnosis confirmed through NS1 dengue and IgM and IgG anti-dengue rapid tests. Blood was drawn from each patient upon enrollment and subsequently at two time points: the critical phase and the day of hospital discharge. We applied the WHO (1997) categories for the clinical phases of dengue. During the critical phase, we looked for pleural effusion, ascites, or increased gallbladder thickness of more than 3 mm, which are the gold standards for determining plasma leakage, using mobile ultrasound.[11] Following a previous study,[12] we performed RT-PCR and serotyping on acute specimens.

The microhematocrit method[13] was used to measure the amount of Ht (in %) in the patients' EDTA blood samples within 2 hours of collection. Hb in the same EDTA blood samples was measured (in g/dl) using the HemoCue® portable photometer (Hb-POCT) following the manual's instructions.[13] The HemoCue® Hb 201 (HemoCue AB,Ängelholm, Sweden) comprises disposable micro cuvettes that contain a dry form of the reagent and a photometer designed for a single purpose. The description of the reaction occurred within the test and operational procedure of the test was in accordance with the operational manual.[13] Examination of Ht and Hb-POCT was conducted on the first day of hospitalization and then daily until the recovery phase. Ethical approval for this study was obtained from the ethics committee of the Faculty of Medicine at Padjadjaran University (approval no. 04/UN6.C2.1.2/KEPK/PN/2014). All procedures followed were in accordance with the ethical standards of the Helsinki Declaration. All study participants provided written informed consent. In patients aged 14–18 years, a parent or guardian provided informed consent with written assent by the child.

Distribution similarities between Hb-POCT and HT were first determined using Pearson's correlation coefficient and a simple linear regression analysis. Next, we ascertained the grouping of patient as dengue hemorrhagic fever (DHF) or dengue fever (DF) using ultrasonographic findings of plasma leakage. The DHF versus DF groups were further described and utilized as the gold standard for determining accuracy of Hb-POCT and HT tests.

We applied three definitions to determine hemoconcentration obtained through Hb-POCT and Ht testing. First, we used the test results at the time of hospital admission and decided that hemoconcentration was present if Hb-POCT and Ht show higher value than the optimal cutoff point as determined by their receiver operating characteristics (ROCs) with the Youden's index.[14,15] Second, we used the delta values of Hb-POCT and HT gathered during admission and follow-up tests. These values were calculated by subtracting the highest to the lowest value divided by the lowest values (presented in percentages) of Hb-POCT and HT observed during hospitalization. Hemoconcentration is determined when the delta value exceeds the optimal cutoff point as informed by ROC analysis.[15] Third, we deemed that hemoconcentration had occurred if the delta value of each parameter increased ≥20% as defined in the WHO dengue guidelines.[5] We then calculated the sensitivity and specificity of Hb POCT and Ht based on each of these three definitions of hemoconcentration compared to DHF and DF category as the gold standard.[13] Lastly, we analyzed the concordance of Hb-POCT vs Ht sensitivity and specificity result using Cohen's Kappa values.

We calculated median and interquartile range (IQR) values for quantitative variables with a non-normal distribution and arithmetic means and standard deviations for quantitative variables with a normal distribution, which were tested with the Shapiro–Wilk test. A *t*-test was performed for independent samples with equal or unequal variations relating to the Levene test result to compare two means. Frequencies of categorical data were compared with the Chi-square test. All analyses were performed using the IBM SPSS Statistics (Version 23) predictive analytics software. A *p*-value <0.05 was considered statistically significant.

Results

Table 1 shows the characteristics of 48 subjects who enrolled in the study. The majority of the subjects were male (65%), and their mean age was 26.4 years (95% confidence interval (CI): 22.8–30 years). Dengue was confirmed through positive NS1 rapid testing in 41 cases (89%) and/or IgM in 18 (60%) and/or IgG rapid testing in 16 cases (53%) of the 30 cases tested. Of seven cases with negative results or no NS1 testing, three dengue cases had RT-PCR positive results, and four had suggestive clinical characteristics (Table 1).

We found gallbladder thickening in all 33 patients diagnosed with DHF; of these patients, 3 (9.1%) also had ascites, 4 (12.1%) had pleural effusion, and 7 (21.2%) had both ascites and pleural effusion. Patients with DHF have significantly higher Hb-POCT and Ht levels at admission, and their delta values between the highest and lowest results during their hospitalization (Table 1). Linear regression analysis shows strong correlation ($R^2 = 0.845$, $p < 0.001$) between Hb-POCT and Ht test results at admission with final Pearson's coefficient of Ht = 8.94 + 2.3 Hb-POCT (Figure 1). This result points out to the similarities of performance of Hb-POCT vs Ht in various operational values that they are supposed to tests.

Table 1 Characteristics of Observed Subjects with Plasma Leakage (Dengue Hemorrhagic Fever) versus Those with No Plasma Leakage (Dengue Fever)

Variable (n = 48)	DHF (n = 33)	DF (n = 15)	*p*-value
Sex, male	72.7%	46.7%	0.08[c]
Age (years), median (IQR)	21 (18–33)	20 (15–35)	0.68[b]
Fever day on admission, median (IQR)	4 (3–5)	4 (3–4)	0.53[b]
Dengue confirmation test			
Positive NS1 Dengue Rapid Test	28/32	13/14	1.00[d]
Positive IgM Dengue Rapid Test	14/23	4/7	
Positive IgG Dengue Rapid Test	13/23	3/7	
Dengue Infecting Serotype			
DEN1	2	0	0.40[c]
DEN2	3	1	
DEN3	11	9	
DEN4	3	1	
Inconclusive PCR	14	3	
Plasma leakage (by ultrasound)			
Pleural effusion	11	0	
Ascites	10	0	
Gallbladder thickening	33	0	
Hb POCT (gr/dl) (mean ± SD)			
On admission	15.6 ± 1.6	14.4 ± 1.3	0.01[a]
Increased Hb (Delta) (%)	11.77 ± 9.1	7.96 ± 5.1	0.13[a]
Ht (%) (Mean ± SD)			
On admission	45.1 ± 4.2	41.5 ± 2.7	0.001[a]
Increased Ht (Delta) (%)	12.18 ± 8.39	7.66 ± 4.93	0.02[a]

Notes: Categorical data were presented as percentages. Continuous variables were presented using means (with standard deviations). Differences between groups were analyzed using [a]Unpaired *t*-test, [b]Mann–Whitney test, [c]Chi Square test, [d]Fisher Exact test. A *p*-value <0.05 was consider significant.

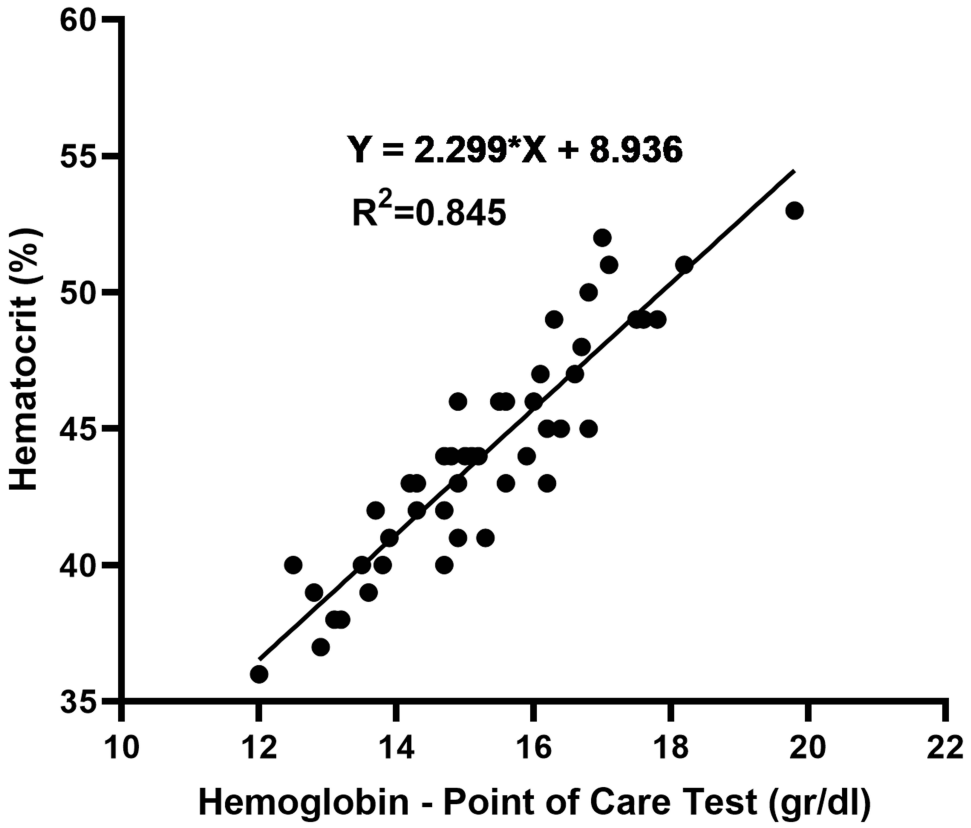

Figure 1 The correlation between hemoglobin-point-of-care testing and hematocrit tests calculated using a simple linear regression.

The AUC of Hb-POCT at admission was slightly lower than that of Ht (0.712 vs 0.746 respectively; Table 2), but the difference was not significant ($p = 0.365$). Our analysis revealed that the optimal cutoff points for Hb-POCT and Ht at admission were, respectively, 14.9 gr/dl and 43%. According to these cutoff points, at the time of admission, Ht measurement had slightly better sensitivity (66.7% vs 63.6%) and slightly better specificity (80.0% vs 73.3%) in detecting plasma leakage compared with Hb POCT, but the difference was not significant. These non-significant results indicated that Hb-POCT performed similarly to Ht testing in detecting plasma leakage. With this level of sensitivity, we missed 12 out of 33 (36%) cases with Hb-POCT and 11 (33%) with Ht test for detecting DHF cases (Table 2).

The AUC values for delta Hb POCT and Ht were 0.615 and 0.643, respectively, in the ROC analysis ($p = 0.705$). The optimal cutoff points for delta Hb-POCT and HT for determining plasma leakage were 10.3% and 10.8%, respectively. These cutoff points indicated slightly lower values for sensitivity and specificity of delta Hb-POCT measurements compared with delta Ht (45.5% vs 48.5% and 80.0% vs 86.7%, respectively), but these differences were not statistically significant. With this level of sensitivity, we missed 18 (54%) cases when using Hb-POCT and 17 (51%) with Ht test for detecting cases with plasma leakage (Table 2).

Using the WHO suggested cutoff values of ≥20% to confirm plasma leakage[5] resulted in a very low sensitivity for detection of plasma leakage. The Hb-POCT shows slightly higher sensitivity of 18.2% relative to Ht of 15.2% but without statistical significance. With this low level of sensitivity, we can only diagnosed 6 (18%) cases when using Hb-POCT and 5 (15%) with Ht test as having DHF (Table 2). Similar accuracies of Hb-POCT and Ht in determining plasma leakage show that these tests have comparable performance.

We also found that Cohen's Kappa values for Hb-POCT and Ht were highly consonant using the optimal cutoff points at the time of admission with 87.5% agreement (Kappa value of 0.75; 95% CI: 0.56–0.94). Determination of plasma leakage of Hb-POCT versus Ht using delta values at optimal cutoff points showed 81.3% agreement and a Kappa value of 0.60 (95% CI: 0.37–0.84). A delta value of ≥20% resulted in 97.9% agreement and a Kappa value of 0.90 (95% CI:

Table 2 Accuracy of Various Methods for Assessing Hemoconcentration Using Optimal and 20% Increase Cutoff Points to Detect Plasma Leakage

	Cutoff	DHF N = 33	DF N = 15	Sensitivity (%)	Specificity (%)	AUC (95% CI for AUC)
At admission with optimal cutoff						
Hemoglobin-POCT	>14.9 gr/dl	21	4	63.6	73.3	0.712 (0.563–0.834)
	≤14.9 gr/dl	12	11			
Hematocrit	>43%	22	3	66.7	80.0	0.746 (0.600–0.861)
	≤43%	11	12			
Delta value using optimal cutoff						
Hemoglobin-POCT	>10.3%	15	3	45.5	80.0	0.615 (0.464–0.752)
	≤10.3%	18	12			
Hematocrit	>10.8%	16	2	48.5	86.7	0.643 (0.492–0.776)
	≤10.8%	17	13			
Delta value using 20% cutoff *						
Hemoglobin-POCT	≥20%	6	0	18.2	100	Not applicable
	<20%	27	15			
Hematocrit	≥20%	5	0	15.2	100	Not applicable
	<20%	28	15			

Note: *WHO criteria 1997.
Abbreviations: DHF, dengue hemorrhagic fever; DF, dengue fever; AUC, area under curve; CI, confidence interval.

0.70–1.00). These high Kappa values and agreement levels indicated similar performances of Hb-POCT and Ht testing in determining plasma leakage.

Discussion

In light of our findings, we concluded that the accuracy of Hb-POCT is similar and not inferior to that of Ht testing for determining hemoconcentration in dengue patients. To the best of our knowledge, this is the first study to demonstrate the value of a handheld hemoglobinometer in diagnosing plasma leakage in dengue patients with reference to ultrasound findings. HemoCue® is one of the most widely used handheld hemoglobinometers for measuring Hb in resource-limited settings. It is relatively inexpensive, portable, does not require a cold chain, and produces a result within a minute.[16] Moreover, it is widely used within mother–child health programs and at blood donation centers.[7,8]

The characteristics of subjects in the DF and DHF groups were similar, although there were more males and older subjects in the DHF group. Dengue infections were confirmed mostly by positive NS1 and PCR, while in four cases, dengue diagnoses were made based on suggestive clinical manifestation. The proportion of patients with plasma leakage, as determined through ultrasound examinations, was high, probably because of the broad definition that we used to define plasma leakage. Hb and Ht measurements were generally less sensitive than ultrasound for detecting plasma leakage.[11] Our findings endorse the value of ultrasonography applications within clinical and research settings.

Although they demonstrate less sensitivity compared with ultrasound, Hb-POCT and Ht testing can potentially be used to detect hemoconcentration as a proxy for plasma leakage. Our results supported those of a previous study that revealed a strong correlation between Hb and Ht measurements.[6] Thus, lower Hb levels are linked to lower concentrations of red blood cells and plasma leakage. Our Kappa values and correlation coefficients showed that Hb measurements obtained using a handheld POCT device were similar to those obtained through Ht testing. Using the suggested cutoff point, we found that the sensitivity of Hb-POCT and Ht values at the time of admission (63.6% and 66.7%, respectively) were more sensitive than the delta values between the highest to the lowest values obtained during hospitalization (45.5% and 48.5%, respectively), with no significant difference in specificity. We believe that Hb-POCT and Ht measurements at the time of admission can be reliably used for making referral decisions or providing fluid treatment. The sensitivity of both Hb-POCT and Ht appeared to be very low when using cutoff values ≥20% increase in value as recommended by the

WHO.[5] Therefore, we suggest that when repetitive measurements of either Hb-POCT or Ht consistently increase even if it has not achieved >20%, health-care workers should assume that hemoconcentration is already present.

Our results support those of previous studies indicating that Hb is adequate for detecting hemoconcentration.[17–19] A previous study found that measuring Hb with HemoCue® using venous blood yielded a high level of accuracy compared with the accuracy of results obtained with a hematology analyzer.[8,9] Another study also proposed calculating the increase in delta Hb values to detect plasma leakage.[17] Beyond the good performance of Hb to detect hemoconcentration as reported, our study can also conclude that determination of hemoconcentration of Hb-POCT or Ht at admission compared to the optimal cutoffs did show better sensitivity than using delta values to detect plasma leakage.

This study had several limitations. First, although we recruited subjects from three hospitals to improve generalizability, the total sample size was small, which meant that we could not adjust our results relative to various confounders, such as sex and age. Additionally, we did not blind the Ultrasound process of determining plasma leakage because the operators were the clinician attending the patients. To compensate for this limitation, Hb-POCT and Ht results used in this analysis were only informed to the research team at the end of the study process.

Conclusion

Our study showed that the accuracy of the Hb-POCT measurement was similar to that of Ht in detecting plasma leakage in dengue-infected subjects. Therefore, we recommend that this test be prioritized along with the NS1 dengue test for first-level care at clinics where Ht and a complete hematology analyzer are unavailable. Hb-POCT is also easy, convenient and provides a quick result which is essential in the management decision of Dengue patients. Because of the low sensitivity of both tests in detecting plasma leakage, we recommend using a lower cut-off point for Hb-POCT and Ht to obtain early indications of hemoconcentration in dengue patients. Further studies are needed to determine appropriate cutoff points differentiated by sex.

Acknowledgments

We acknowledge the contributions of Dr. Haneng Marissangan SpPD and Dr. Any Yuliani SpPK M.Kes, and the staff at Dr. M. Salamun Hospital in Bandung for their support. We also thank personnel at the Immunology Laboratory, Faculty of Medicine, at Universitas Padjadjaran, Siti Rasnawati Mony, and Hofiya Djauhari for their technical assistance. We thank staff of the HEALTH-I Project for providing input and assistance during analysis and writing of this article.

Disclosure

The authors declare that they have no conflicts of interest in this work.

References

1. World Health Organization. Global strategy for dengue prevention and control 2012–2020. Switzerland: WHO Library Cataloguing-in-Publication Data; 2012. Available from: https://apps.who.int/iris/bitstream/handle/10665/75303/9789241504034_eng.pdf. Accessed June 26, 2021.
2. Guzman MG, Harris E. Dengue. *Lancet*. 2015;385(9966):453–465. doi:10.1016/S0140-6736(14)60572-9
3. Gubler DJ. Dengue/dengue haemorrhagic fever: history and current status. *Novartis Found Symp*. 2006;277:3–16;discussion 16–22, 71–3, 251–3. doi:10.1002/0470058005.ch2
4. Halstead SB. Dengue. *Lancet*. 2007;370(9599):1644–1652. doi:10.1016/S0140-6736(07)61687-0
5. World Health Organization/Regional Office of South East Asia. Comprehensive guidelines for prevention and control of dengue and dengue haemorrhagic fever. Revised and expanded edition; 2011. Available from: https://apps.who.int/iris/handle/10665/204894. Accessed August 18, 2021.
6. Nijboer JMM, van der Horst ICC, Hendriks HGD, Duis HJ, Nijsten MWN. Myth or reality: hematocrit and hemoglobin differ in trauma. *J Trauma*. 2007;62(5):1310–1312. doi:10.1097/TA.0b013e3180341f54
7. Nkrumah B, Nguah SB, Sarpong N, et al. Hemoglobin estimation by the HemoCue® portable hemoglobin photometer in a resource poor setting. *BMC Clin Pathol*. 2011;11(1):5. doi:10.1186/1472-6890-11-5
8. Sanchis-Gomar F, Cortell-Ballester J, Pareja-Galeano H, Banfi G, Lippi G. Hemoglobin point-of-care testing: the HemoCue system. *J Lab Autom*. 2013;18(3):198–205. doi:10.1177/2211068212457560
9. Jain A, Chowdhury N. Comparison of the accuracy of capillary hemoglobin estimation and venous hemoglobin estimation by two models of HemoCue against automated cell counter hemoglobin measurement. *Asian J Transfus Sci*. 2020;14(1):49–53. doi:10.4103/ajts.AJTS_93_17
10. Jain A, Chowdhury N, Jain S. Intra- and inter-model reliability of Hemocue Hb 201+ and HemoCue Hb 301 devices. *Asian J Transfus Sci*. 2018;12:123–126. doi:10.4103/ajts.AJTS_119_17

11. Michels M, Sumardi U, de Mast Q, et al. The predictive diagnostic value of serial daily bedside ultrasonography for severe dengue in Indonesian adults. *PLoS Negl Trop Dis*. 2016;7(6):e2277. doi:10.1371/journal.pntd.0002277
12. Lanciotti RS, Calisher CH, Gubler DJ, Chang GJ, Vorndam AV. Rapid detection and typing of dengue viruses from clinical samples by using reverse transcriptase-polymerase chain reaction. *J Clin Microbiol*. 1992;30(3):545–551. doi:10.1128/jcm.30.3.545-551.1992
13. The HemoCue® Hb 201+ operating manual, manufactured by HemoCue AB, Ängelholm, Sweden. Available from: https://www.hemocue.us/wp-content/uploads/2020/06/Manual_HB_201.pdf. Accessed November 28, 2021.
14. Fluss R, Faraggi D, Reiser B. Estimation of the Youden Index and its associated cut-off point. *Biom J*. 2005;47(4):458–472. doi:10.1002/bimj.200410135
15. De Long ER, De Long DM, Clarke-Pearson DL. Comparing the areas under two or more correlated receiver operating characteristic curves: a nonparametric approach. *Biometrics*. 1988;44(3):837–845. doi:10.2307/2531595
16. Pullum TW, Collison DK, Namaste S. Hemoglobin data in DHS surveys: intrinsic variation and measurement error, 2017. Available from: https://dhsprogram.com/pubs/pdf/MR18/MR18.Pdf. Accessed August 18, 2021.
17. Ralapanawa U, Alawattegama ATM, Gunrathne M, Tennakoon S, Kularatne SAM, Jayalath T. Value of peripheral blood count for dengue severity prediction. *BMC Res Notes*. 2018;11(1):400. doi:10.1186/s13104-018-3505-4
18. Premaratna R, Ragupathy A, Miththinda JKND, de Silva HJ. Timing, predictors, and progress of third space fluid accumulation during preliminary phase fluid resuscitation in adult patients with dengue. *Int J Infect Dis*. 2013;17(7):e505–e509. doi:10.1016/j.ijid.2012.12.021
19. Wisanuvej K, Boonyawat K, Savetamornkul C, et al. Comparison between blood hemoglobin concentration determined by point-of-care device and complete blood count in adult patients with dengue. *PLoS Negl Trop Dis*. 2021;15(8):e0009692. doi:10.1371/journal.pntd.0009692